Antimicrobial PLA and Calcium Phosphate Coatings and Thin Film Composites for Implants Applications

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the degree of

Doctor of Philosophy (Science)

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CERTIFICATE OF ORIGINAL AUTHORSHIP

I, [Ipek Karacan Soylu] declare that this thesis is submitted in fulfilment of the requirements for the award of [Doctor of Philosophy], in the [Faculty of Science / School of Life Science] at the University of Technology Sydney.

This thesis is wholly my own work unless otherwise reference or acknowledged. In addition, I certify that all information sources and literature used are indicated in the thesis.

This document has not been submitted for qualifications at any other academic institution.

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Abstract

Implant-related infections after the insertion of biomedical implants are still prevalent, and the current treatment methodology requires the use of large doses of antibiotics systemically. Large doses can lead to a number of side effects including antibiotic resistance and adverse effects on the other organs. These infections delay healing, worsen functional outcome and incur significant socio-economic costs. Bone implant related infections, surgery site infections (SSI), and osteomyelitis remain the most challenging clinical problems faced. This thesis aimed to develop a novel implant coating system based on a 'Local and Controlled Antibiotic Delivery System with Biodegradable Polymeric Thin Film Composite Coating' for metallic bone implants.

The specific aim of this study was to design a novel multi-functional and antibacterial coating for implants as the drug delivery systems to prevent post-operative complications and osteomyelitis. The main challenge was to obtain the perfect design and the selection of appropriate biomaterials; implantable device with antibacterial, biocompatible and bioactive properties. Therefore, gentamicin antibiotic (Gm) and Gm loaded coralline hydroxyapatite (HAp) particles were incorporated into a poly-lactic acid (PLA) matrix as the main biocomposite. A number of systems were produced, characterized and tested, which included PLA, PLA-Gm mixture, and a PLA-Gm-(HAp-Gm) biocomposite.

The coral skeleton ($CaCO_3$) was converted to HAp by using the hydrothermal conversion method. These microspheres were loaded with Gm and HAp-Gm particles and incorporated within the PLA thin film composites. Coralline-HAp possesses a unique nano- and meso-porous structure and can be used as a drug carrier for the sustained release of antibiotics on metallic bone implants. While the physiochemical characterizations of the PLA biocomposite coating were evaluated, their Gm release profile were analyzed by the continuous dissolution method. The bioactivity and biocompatibility of the design was tested on Adipose-derived stem cells using *in vitro* studies. The antibacterial activity and biofilm formation behaviour were also analyzed with *S. aureus* and *S. epidemidis*.

Different Gm concentrations (5%, 10%, 15%, 20% and 30% [w/w]) were incorporated into the PLA biocomposites and were found to be highly effective on the inhibition of *S. aureus* and *S. epidermidis* growth at the planktonic stage. At the biofilm formation stage of S. *aureus*, the significant reduction of bacterial attachment and the increasing of dead microcolonies were observed for even the lowest 5% (w/w) PLA-Gm-(HAp-Gm) coated samples. This research showed that the biodegradable and antibacterial PLA biocomposite coatings design has high potential as a viable alternative method for existing clinical applications on many metallic orthopedic and maxillofacial bone implants.

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initiated). Absorbance was measured at 600 nm for twenty-four hours. Statistical analysis performed using two-way ANOVA with Tukey's post-test. n=3. Error bars denote SD. **p<0.01, ****p<0.0001

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Figure 6. 6 Graph represents the percentage of DMOA-positive (live) *S. aureus* following twenty-four hours of co-culture with the controls (A) (as indicated), and (B) PLA and PLA-Gm-(HAp-Gm) thin film coated Ti6Al4V discs (5% to 30% w/w Gm concentrations). The analysis was performed using ImageJ software. Statistical analysis performed using one-way ANOVA with Tukey's post-test. n=3 (15 images per sample). Error bars denote SEM. *p<0.05, **p<0.01

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Figure 7. 6 Graph represents the comparison of (A) the cell numbers, (B) total cell area %, (C) single cell area mm^2, and (D) circularity for PLA-Gm-(HAp-Gm) thin film coated Ti6Al4V discs (5% to 30% w/w Gm concentrations). The analysis was performed using ImageJ software. Statistical analysis performed using one-way ANOVA with Tukey's post-test. n=3. Error bars denote SEM. P values: *p<0.04, **p<0.008, **** p<0.0001

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2. Mutations of a conserved tryptophan residue of the TEM 1 ß lactamase, (2015), The 29th Annual Meeting of the Protein Society, Barcelona, Spain – Poster Presentation
3. Antibiotic containing poly lactic acid/hydroxyapatite biocomposite coatings for dental implant applications, (2017), Bioceramics 29 - Toulouse, France – Oral and Poster Presentation
4. Development of PLA Biocomposite Coatings as Drug Delivery System for Dental Implants, (2018) The Australasian Society for Biomaterials and Tissue Engineering community (ASBTE) Meeting - Oral Presentation
5. Development and Testing of a New Antibiotic Loaded Biodegradable Biocomposite Coating on Ti6Al4V Metallic Implants, (2018), Australian Society for Medical Research Annual Meeting – Poster Presentation
6. Multifunctional-Dual Drug Delivery PLA Coating with HAp for Bone Implants, (2018), Bioceramics30, Nagoya, Japan- Oral and Poster Presentations